

(19) World Intellectual Property
Organization
International Bureau



(43) International Publication Date
9 September 2005 (09.09.2005)

PCT

(10) International Publication Number
WO 2005/083068 A2

(51) International Patent Classification⁷: C12N 9/12

(21) International Application Number:
PCT/IB2005/000734

(22) International Filing Date: 25 February 2005 (25.02.2005)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
10/787,219 27 February 2004 (27.02.2004) US

(71) Applicants (for all designated States except US): **INSTITUT PASTEUR** [FR/FR]; 25-28, rue du Docteur Roux, F-75015 Paris (FR). **CENTRE NATIONAL DE LA RECHERCHE SCIENTIFIQUE (CNRS)** [FR/FR]; 3, rue Michel Ange, F-75016 Paris (FR).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **JESTIN, Jean-Luc** [FR/FR]; 42, rue des Gravilliers, F-75003 Paris (FR). **VICHIER-GUERRE, Sophie** [FR/FR]; 1, Allée de Savoie, F-78170 La Celle Saint-Cloud (FR). **FERRIS, Stéphane** [FR/FR]; 4, Place des Eglantines, F-91540 Mennecy (FR).

(74) Agents: **MARTIN, Jean-Jacques** et al.; Cabinet Regimbeau, 20, rue de Chazelles, Cedex 17, F-75847 Paris (FR).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

- without international search report and to be republished upon receipt of that report
- with (an) indication(s) in relation to deposited biological material furnished under Rule 13bis separately from the description

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHODS FOR OBTAINING THERMOSTABLE ENZYMES, DNA POLYMERASE I VARIANTS FROM *THERMUS AQUATICUS* HAVING NEW CATALYTIC ACTIVITIES, METHODS FOR OBTAINING THE SAME, AND APPLICATIONS TO THE SAME

(57) Abstract: The present invention provides a method for obtaining thermostable enzymes. The present invention also provides variants of DNA polymerase I from *Thermus aquaticus*. The present invention further provides methods of identifying mutant DNA polymerases having enhanced catalytic activity. The present invention also provides polynucleotides, expression systems, and host cells encoding the mutant DNA polymerases. Still further, the present invention provides a method to carry out reverse transcriptase-polymerase chain reaction (RT-PCR) and kits to facilitate the same.



WO 2005/083068 A2